

Appendix C

Tobamovirus RNA dependent RNA polymerase

A three-dimensional structure of a RNA-dependent RNA polymerase, that encoded by poliovirus, has been reported by Hansen et al. The alignment used for tree construction is enhanced below by the amino acid sequence of the poliovirus RdRP aligned to the tobamoviral RdRp's. A 3D line is provided that identifies the helices (upper case Roman) and beta-structure elements (numerals) described in that report. The footnote line refers to notes at the bottom that relate to functional or structural comments.

	1
<u>TMV-vul</u>	--MQFYFDKCLPGNSTMMNNFD---AVTMRLTDISLNVKDCILDMSKSVA
<u>TMV-Rak</u>	--MQFYFDKCLPGNSTMMNNFD---AVTMRLTDISLNVKDCILDMSKSVA
<u>TMV-WANG</u>	--MQFYFDKCLPGNSTMMNNFD---AVTMRLTDISLNVKDCILDMSKSVA
<u>TOMV-L</u>	--MQFYFDKCLPGNSTILNNYD---AVTMKLTDISLNVKDCILDMSKSVA
<u>TMV-KR</u>	--MQFYFDKCLPGNSTMMNNFD---AVTMRLTDISLNVKDCILDMSKSVA
<u>PMMV</u>	--MQYYFDKCLPGNSTILNEYD---AVTMQIRENSLNVKDCVLDMSKSV
<u>TMGMV</u>	PDLQFYFDVCLPGNSTILNKYD---AVTMRLRDNSLNVKDCVLDKFSKSI
<u>ORSV</u>	--MQFYDITLLPGNSTILNEYD---AVTMNLRENNLNVKDCITDFSKSV
<u>TVCV</u>	--MQFYDALLPGNSTILNEFD---AVTMNLRDISLNVKDCRIDFSKSVQ
<u>CR-TMV</u>	--MQFYDITLLPGNSTILNEYD---AVTMNLRDISLNVKDCRIDFSKSVQ
<u>RMV-SH</u>	--MQFYDITLLPGNSTILNEFD---AVTMNLRDISLNVKDCRIDFSKSVQ
<u>CRMV</u>	--MQFYDITLLPGNSTILNEFD---AVTMNLRDISLNVKDCRIDFSKSVQ
<u>TMV-CG</u>	--MQFYDITLLPGNSTILNEFD---AVTMNLRDISLNVKDCRIDFSKSVQ
<u>TMV-OB</u>	QDLQFYFDKCLPGNSTVLNEFD---AVTMNCSDISLNVKDCVLDKFSKSV
<u>CGMMV</u>	--MQEFYDRCLPGNSFVLNDFD---AVTMRLRDNEFNLQPCRLTSLNLD
<u>CGMMV-W</u>	TDMQEFYDRCLPGNSFVLNDFD---AVTMRLRDNEFNLQPCRLTSLNLD
<u>CFMMV</u>	--MQNFYDACLPNGSFVLNDYD---SVTMRLVDNEINLQPCRLTSLKADP
<u>YCGMMV</u>	TDMQSFYDACLPNGSFVLNDYD---SVTMRLADNEFNLQPCRLTSLKADP
<u>SHMV</u>	--LQYFYDSWLPNGSFVQNNHD---QWSIISSDINLHSEAVRLDMNKRH-
<u>POLIORDRP</u>	GEIQWMPRSKEVGYPINAPSKTKLEPSAFHY-VFEGVKEPAVLTKNDRP
3D	-----
Footnote	11 1
TMV VUL	AP---KDQIK-PLIPMVRTAAEMPRQTGLLENLVAMIKRNFNAPELSGII
TMV-RAK	AP---KDQIK-PLIPMVRTAAEMPRQTGLLENLVAMIKRNFNAPELSGII
TMV WANG	AP---KDQIK-PLIPMVRTAAEMPRQTGLLENLVAMIKRNFNAPELSGII
TOMV L	AP---KDVKP-TLIPMVRTAAEMPRQTGLLENLVAMIKRNFNSPELSGVV
TMV-KR	AP---KDQIK-PLIPMVRTAAEMPRQTGLLENLVAMIKRNFNAPELSGII
PMMV	LP---RESET-TLKPVIRTAAEKPRKPGLLENLVAMIKRNFNSPELVGVV
TMGMV SPA	MP---KEVKP-CLEPVLRTAAEPRAAGLLENLVAMIKRNFNAPDLTGTI
ORSV	VP---RQEE-FFTPVIRTAAPERPRAGLLENLVAMIKRNFNSPDLTGIL
TVCV	LP---KEQPI-FLKPKIRTAAMPRTAGLLENLVAMIKRNMNAPDLTGTI
CR-TMV	LP---KEQPI-FLKPKIRTAAMPRTAGLLENLVAMIKRNMNAPDLTGTI
RMV-SH	VP---KERPV-FMKPKLRTAAEMPRTAGLLENLVAMIKRNMNAPDLTGTI
CRMV	VP---KERPV-FMKPKLRTAAEMPRTAGLLENLVAMIKRNMNAPDLTGTI
TMV-CG	LP---RERPI-FMKPKLRTAAEMPRTAGLLENLVAMIKRNMNAPDLTGTI
TMV OB	LP---RDNTKVPMTVPVIRTAAPERPRSQGLLENLVAMIKRNFNSPELSGT
CGMMV	VPALIKNEAQNFLIPVLRACERPRIPGLLENLVAMIKRNMNTPDLAGTV
CGMMV-W	VPALVKSEAQNFLIPVLRACERPRIPGLLENLVAMIKRNMNTPDLAGTV
CFMMV	VTESLKMEKKEFLIPLGKTATERPRIPGLLENLIAIVKRNFNTPDLAGSL
YCGMMV	VAESIKLERKNIDKLDKTATERPRIPGFLENLVAIVKRNFNTPDLAGVL
SHMV	----IPRTKGEFLRPLLNTAVEPPRIIPGLLENLLALIKRNFNAPDLAGQL
POLIORDRP	L----K-TNFEEAIFSKYVGNKITEVDEHMKEAVDHYAGQLMSLDINTEQ
3D	-----AAAAAAAAAAAAAAAA-----
Footnote	
TMV VUL	DIENTASLVVDKFFDSYLLKE-----KRKPNKNVSLFSRESLNRWLEKQE
TMV-RAK	DIENTASLVVDKFFDSYLLKE-----KRKPNKNVSLFSRESLNRWLEKQE
TMV WANG	DIENTASLVVDKFFDSYLLKE-----KRKPNKNVSLFSRESLNRWLEKQE
TOMV L	DIENTASLVVDKFFDSYLLKE-----KRKPNKNVSLFSRESLNRWLEKQE
TMV-KR	DIENTASLVVDKFFDSYLLKE-----KRKPNKNVSLFSRESLNRWLEKQE

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PMMV      DIEDTASLVVDKFFDAYLIKE-----KKKP-KNIPLLSRASLERWIEKQE
TMGMV SPA DIESTASVVVDKFFDSYFIKK-----EKYTKNIAGVMTKDSMMRWLENRK
ORSV      DIEDTAELVVNKFWDAYIIDE-LSGGNVTPM-----TSDAFHRWMAKQE
TVCV      DIEDTASLVVEKFWDSDYVDKE-FSGTNEMTM-----TRESFSRWLSKQE
CR-TMV    DIEDTASLVVEKFWDSDYIDKE-FSGTNEMTM-----TRESFSRWLSKQE
RMV-SH    DIEDTASLVVEKFWDAYVVKE-FSGTDGMAM-----TRESFSRWLSKQE
CRMV      DIEDTASLVVEKFWDAYVVKE-FSGTDGMAM-----TRESFSRWLSKQE
TMV-CG    DIEDTASLVVEKFWDAYVVKE-FSGTDGMAM-----TRESFSRWLSKQE
TMV OB    DMENTASVADRFFDSYFLKDKLSGCSLGDSSGKNIIDRQALIRWMEKQE
CGMMV     DITNMSISIVDNFFSSFVRDE-V---LLDHLDCVRASSIQSFSDWFSCQP
CGMMV-W   DITNMSISIVDNFFSSFVRDE-V---LLDHLDCVRASSIQSFSDWFSCQP
CFMMV     DISSISKGVVDNFFSTFLRDE-Q---LADHLCKVRSLSEFSFAWFDNQS
YCGMMV    DIDTISKSVVDNFFTTFLRDE-Q---LSDHLVRVRSCSLESFSAWFHNQA
SHMV      DYDFLSRKVCDGFFGKLLPPD-VEASELLRLPVDHMYSVQNFDDWLNKQE
POLIORDRP MCLEDAMYGTGDLALDLSTS-----AGYPYVAMGKKKRDILN-----
3D

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Footnote

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TMV VUL    QVTIGQLADFDVFDLPAVDQYR-HMYKAQPKQKLDTSIQTEYPA-LQTIV
TMV-RAK    RVTIGQLADFDVFDLPAVDQYR-HMIKAQPKQKLDTSIQTEYPA-LQTIV
TMV WANG   QVTIGQLADFDVFDLPAVDQYR-HMIKAQPKQKLDTSIQTEYPA-LQTIV
TOMV L     QVTIGQLADFDVFDLPAVDQYR-HMIKAQPKQKLDLSIQTEYPA-LQTIV
TMV-KR     QVTIGQLADFDVFDLPAVDQYR-HMIKAQPKQKLDTSIQTEYPA-LQTIV
PMMV       KSTIGQLADFDLPAVDQYR-HMIKQPKQRLDLSIQTEYPA-LQTIV
TMGMV SPA  EVLLDDLANYNFTDLPAIDQYK-HMIKAQPKQKLDLSIQNEYPA-LQTIV
ORSV       KSTIRQLADFDVFDLPAIDQYK-HMIKAQPKQKLDLSPQDEYAA-LQTIV
TVCV       SSTVGQLADFNFDLPAVDEYK-HMIKSQPKQKLDLSIQDEYPA-LQTIV
CR-TMV     SSTVGQLADFNFDLPAVDEYK-HMIKSQPKQKLDLSIQDEYPA-LQTIV
RMV-SH     SSTVGQLADFNFDLPAVDEYK-HMIKSQPKQKLDLSIQDEYPA-LQTIV
CRMV       SSTVGQLADFNFDLPAVDEYK-HMIKSQPKQKLDLSIQDEYPA-LQTIV
TMV-CG     SSTVGQLADFNFDLPAVDEYK-HMIKSQPKQKLDLSIQDEYPA-LQTIV
TMV OB     KSTIGQLADYDFVFDLPAIDQYR-HIISQPKQKLDLSIQSEYPS-LQTIV
CGMMV      TSAVGQLANFNFDLPAFDTYM-HMIKRQPKSRDLSIQSEYPA-LQTIV
CGMMV-W    TSAVGQLANFNFDLPAFDTYM-HMIKRQPKSRDLSIQSEYPA-LQTIV
CFMMV      TCALGQLSNFDVFDLPPVDVYN-HMIKRQPKSKLDTSIQSEYPA-LQTIV
YCGMMV     TAAMGQLANFDLPPVDMYT-HMIKRQPKSKLDTSIQSEYPA-LQTIV
SHMV       PGVVGQLANWDHIGMPAADQYR-HMIKRTPKAKLDLSIQSEYPA-LQTIV
POLIORDRP KQTRDTKEMQKLLDTYGINLPLVTYVKDELRSKTKV-EQGKSRLIEASSL
3D

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Footnote

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TMV VUL    YHSKKINAI FGPLFSELTRQLLDSVDSSRFLFFTRKTPAQIEDFFGDLD
TMV-RAK    YHSKKINAI FGPLFSELTRQLLDSVDSSRFLFFTRKTPAQIEDFFGDLD
TMV WANG   YHSKKINAI FGPLFSELTRQLLDSVDSSRFLFFTRKTPAQIEDFFGDLD
TOMV L     YHSKKINAI FGPLFSELTRQLLDSIDSSRFLFFTRKTPAQIEDFFGDLD
TMV-KR     YHSKKINAI FGPLFSELTRQLLDSVDSSRFLFFTRKTPAQIEDFFGDLD
PMMV       YHSKKINAI FGPFVSELTRQLLETIDSSRFMFYTRKTPAQIEEFFSDLDS
TMGMV SPA  YHSKQINGILAG-FSELTRLLEAFDSKKFLFFTRKTPAQIEEFFSDLDS
ORSV       YHSKQINAI FGPLFAELTRQLLERIDSSKFLFYTRKTPAQIEEFFSDLDS
TVCV       YHSKKINAI FGPMFSELTRMLLERIDSSKFLFYTRKTPAQIEEFFSDLDS
CR-TMV     YHSKKINAI FGPMFSELTRMLLERIDSSKFLFYTRKTPAQIEEFFSDLDS
RMV-SH     YHSKKINAI FGPMFSELTRMLLERIDTSKFLFYTRKTPAQIEEFFSDLDS
CRMV       YHSKKINAI FGPMFSELTRMLLETIDTSKFLFYTRKTPAQIEEFFSDLDS
TMV-CG     YHSKKINAI FGPMFSELTRMLLERIDTSKFLFYTRKTPAQIEEFFSDLDS
TMV OB     YHSKKINAI FGPIFSELTRQMLSAIDTSRYLFFTRKTPAQIEEFFSDLDA
CGMMV      YHPKVNAVFGPVFKYLTTKFLSMVDSSKFFFYTRKKPEDLQEFFSDLSS
CGMMV-W    YHPKVNAVFGPVFKYLTTKFLSMVDSSKFFFYTRKKPEDLQEFFSDLSS
CFMMV      YHSKLVNAVFGPVFRYLTSEFLSMVDNSKFFFYTRKLRMICKFLPHFPN
YCGMMV     YHSKLVNAVFGPVFRYLTSEFLSMVDNSKFFFYTRKTPEDLQSFFSTLSA
SHMV       YHSKLVNAVFGPIFSCLETERLLSVVDPLRFKFFTRTPADLEFFFRDMV
POLIORDRP NDSVAMRMAFGNLYAAAFHK-----NPGVITGSAVGCDPDLFWSKIPV
3D        ----BBBBB-CCCCCCCC-----DDDDDD

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Footnote	5	aaaaaa
TMV VUL	IIKGAFCGDDSLLYFPKGCEFPDV-QHSA--NLMWNFEAK-----LFKKQ	
TMV-RAK	IIKGAFCGDDSLLYFPKGCEFPDI-QHSV--NLMWNFEAK-----LFKKQ	
TMV WANG	IIKGAFCGDDSLLYFPKGCEFPDV-QHSA--NLMWNFEAK-----LFKKQ	
TOMV L	LIKGAFCGDDSLLYFPKGCEYPDI-QQAA--NLMWNFEAK-----LFKKQ	
TMV-KR	IIKGAFCGDDSLLYFPKGCEFPDV-QHSA--NLMWNFEAK-----LFKKQ	
PMMV	LIKGAFCGDDSLIYFPKGTDFPDI-QQGA--NLLWNFEAK-----LFRKR	
TMGMV SPA	VIKGAFCGDDSVLYFPKGLDFPDI-QSCA--NLMWNFEAK-----LYRKR	
ORSV	VIKAAFCGDDSIILDI PKGLDLPDI-QSEA--NLMWNFEAK-----LYRKR	
TVCV	VIKAAFCGDDSLIYIPKGLDLPDI-QAGA--NLMWNFEAK-----LFRKK	
CR-TMV	VIKAAFCGDDSLIYIPKGLDLPDI-QAGA--NLMWNFEAK-----LFRKK	
RMV-SH	VIKAAFCGDDSLIYIPKGLDLPDI-QAGA--NLTWNFEAK-----LFRKK	
CRMV	VIKAAFCGDDSLIYIPKGLDLPDI-QAGA--NLTWNFEAK-----LFRKK	
TMV-CG	VIKAAFCGDDSLIYIPKGLDLPDI-QAGA--NLTWNFEAK-----LFRKK	
TMV OB	VIKAAFCGDDSLVLYLPKGCELPNI-QSCA--NLMWNFEAK-----LFKKT	
CGMMV	CFKASFCGDDSLIYLPKGLEYDPI-QATA--NLVWNFEAK-----LFRKK	

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CGMMV-W   CFKASFCGDDSLIYLPKGGLEYPDI-QATA--NLVWNFEAK-----LFRKK
CFMMV     SFKAAFCGDDSLIYMPPNLEYNDI-QSTA--NLVWNFEAK-----LYKKK
YCGMMV    SFKASFCGDDSLIYMPPGLEYPDI-QATA--NLVWNFEAK-----LFKKR
SHMV      VFKAGFCGDDSIYLPRLNLLYPDI-QSVS--NNMWNFEAK-----LFKKL
POLIORDRP HMKMIAYGDDVIASYPHEVDASLLAQSGKDYGLTMTPADKSAIFETVTWE
3D        222222---333333-----IIIIIIIIIIII-4444-----
Footnote          44          9 9
TMV VUL    YGYFCGRYVIHHDRCIVYYDPLKL-ISKLGAKHIKDWEHLEEFRRSLCD
TMV-RAK    YGYFCGRYIIHHDRCIVYYDPLKL-ISKLGAKHIKDWEHLEEFRRSLCD
TMV WANG    YGYFCGRYVIHHDRCIVYYDPLKL-ISKLGAKHIKDWEHLEEFRRSLCD
TOMV L     YGYFCGRYVIHHDRCIVYYDPLKL-ISKLGAKHIKDWDHLEEFRRSLCD
TMV-KR     YGYFCGRYVIHHDRCIVYYDPLKL-ISKLGAKHIKDWEHLEEFRRSLCD
PMMV       YGYFCGRYIIHHDRCIVYYDPLKL-ISKLGAKHIKNREHLEEFRTSLCD
TMGMV SPA  YGYFCGRYIIHHDKGAIVYYDPLKL-ISKLGAKHIKDYDHLEELRVSLCD
ORSV       YGYFCARYIIHHDRCIVYYDPLKL-ISKLGCKHIKSLDHLEEFRRSLCD
TVCV       YGYFCGRYVIHHDRCIVYYDPLKL-ISKLGCKHIRDVVHLEELRESLCD
CR-TMV     YGYFCGRYVIHHDRCIVYYDPLKL-ISKLGCKHIRDVVHLEELRESLCD
RMV-SH     YGYFCGRYVIHHDRCIVYYDPLKL-ISKLGCKHIRDEVHLEELRRSLCD
CRMV       YGYFCGRYVIHHDRCIVYYDPLKL-ISKLGCKHIRDEVHLEELRRSLCD
TMV-CG     YGYFCGRYVIHHDRCIVYYDPLKL-ISKLGCKHIRDEVHLEELRRSLCD
TMV OB     YGYFCGRYVIHHDRCIVYYDPLKI-ISKLGAKHITDKEHLEEFRTSLAD
CGMMV      YGYFCGRYIIHHANGCIVYDPLKL-ISKLGKNSLVGYEHVEEFRTSLD
CGMMV-W    YGYFCGRYIIHHANGCIVYDPLKL-ISKLGKNSLVGYEHVEEFRTSLD
CFMMV      YGYFCGRYIIHHANGCIVYDPLKL-ISKLGKNSLVGYEHVEEFRTSLD
YCGMMV     YGYFCGRYIIHHSNGCIVYDPLKL-ISKLGKNSLVGYEHVEEFRTSLD
SHMV       HGYFCGRYIILNRGRYLRLLPDPLKI-ITKLGCKAIKDWHDLEEFRTSLD
POLIORDRP NVTFLKRFRFADEKYPFLIHPVMPMKEIHESIRWTKDPRNTQDHVRSCL
3D        -555---66666-----7777-JJJJJJJJJJ----KKKKKKKKKKKKKK
Footnote          77          88 8
TMV VUL    VAVSLNN-CAYYTQLDDAVWEVHKTAPPGSFVYKSLVKYLSKVLFRSLF
TMV-RAK    VAVSLNN-CAYYTQLDDAVWEVHKTAPPGSFVYKSLVKYLSKVLFRSLF
TMV WANG    VAVSLNN-CAYYTQLDDAVWEVHKTAPPGSFVYKSLVKYLSKVLFRSLF
TOMV L     VAESLNN-CAYYTQLDDAVGEVHKTAPPGSFVYKSLVKYLSKVLFRSLF
TMV-KR     VAVSLNN-CAYYTQLDDAVWEVHKTAPPGSFVYKSLVKYLSKVLFRSLF
PMMV       VAGSLNN-CAYYTHLNDVGEVIKTAPLGSFVYKSLVKYLSKVLFRSLF
TMGMV SPA  VACSLGNWCLGFPQLNAAIKEVHKTAVNGSFAFNCVNKFLCDKFLFRTLF
ORSV       VSSSLNN-CALFGQLNDAIAEVHKTAVNGSFAFCSIVKYLSKVLFRSLF
TVCV       VASNLNN-CAYFSQLDEAVAEVHKTAVGGSFAFCSIIKYLSKRLFRDLF
CR-TMV     VASNLNN-CAYFSQLDEAVAEVHKTAVGGSFAFCSIIKYLSKRLFRDLF
RMV-SH     VTSNLNN-CAYFSQLDEAVAEVHKTAVGGAFFVYCSIIKYLSKRLFKDLF
CRMV       VTSNLNN-CAYFSQLDEAVAEVHKTAVGGAFFVYCSIIKYLSKRLFKDLF
TMV-CG     VTSNLNN-CAYFSQLDEAVAEVHKTAVGGAFFVYCSIIKYLSKRLFKDLF
TMV OB     VSKSLNN-CAYYAQLDEAVREVHKTAPPGSFVYKSLVKYLSKVLFRSLF
CGMMV      VAHSLFN-GAYFHLDDAIHELFPNAGGCSFVINCLCKYLSKRLFRSLY
CGMMV-W    VAHSLFN-GAYFHLDDAIHELFPNAGGCSFVINCLCKYLSKRLFRSLY
CFMMV      VAKPLFN-AAYFHLDDAIHEYFPSVGGSTFAISSLCKYLSKRLFRSLF
YCGMMV     VAKPLFN-AAYFHLDDAIHEYFPSVGGSSFAINSCLKYLSKRLFRSLF
SHMV       MACEYKN-CFGFDVLESARKESFPAEGCNVAFCAIYKFLSNKYLFRTLF
POLIORDRP LAWHNGE-EEYNKFL-----AKIRSVPIGRALLPEYSTLYRRWL
3D        KKKK--L-LLLLLLL-----LLLL--MMM-----NNNNNNNNNN
Footnote          8          0
TMV VUL    ID
TMV-RAK    IN
TMV WANG    ID
TOMV L     LD
TMV-KR     ID
PMMV       LE
TMGMV SPA  LN
ORSV       ??
TVCV       FV

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CR-TMV	FV
RMV-SH	FV
CRMV	FV
TMV-CG	FV
TMV OB	F?
CGMMV	ID
CGMMV-W	ID
CFMMV	IK
YCGMMV	AK
SHMV	SD
POLIORDRP	DS
3D	NN

Footnote

- Footnotes:
- 1. N-terminal residues interacting with the thumb.
- 4. Three aspartates involved in metal ion binding at the active site.
- 5. Determinant of preference for NTP over dNTP. Could interact with 2' OH of substrate NTP.
- 6. Caps a helical turn
- 7. beta-turn.
- 8. Interaction across interface I
- 9. Interaction across interface I with Arg 456
- 0. See 9.
- a. Extra residues are present between helix H and beta-2 for poliovirus and between helix D and helix C for tobamoviruses. Their sequence similarity (YKGID for polio vs. LDSID for ToMV) suggests that they may serve similar functions in providing part of Interface I.

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